ABSTRACT

Processed cheese food (PCF) is a dairy product prepared by blending dairy ingredients with non-dairy ingredients and heating the blend with agitation to produce a homogeneous product with an extended shelf-life. Emulsifying salts (ES), such as disodium phosphate (DSP) and trisodium citrate, have a critical effect on the emulsification characteristics of casein by sequestering the calcium from the calcium-paracaseinate phosphate complex in natural cheese (NC). Lactose-6-phosphate (LP) is an organic compound produced from lactose that has the potential to function as ES. LP is not approved for use as a substitute for ES in the large-scale production of PC. The objective of this study was to produce PCF with LP instead of DSP. LP was prepared by mixing 1 mol of α-lactose with 0.5 mol of sodium cyclo-triphosphate. The pH of recombined solutions was adjusted using sodium hydroxide to get a pH of 12 to obtain 60.74% LP. The solution was stirred for 3 d at room temperature and then concentrated to 52% total solids (TS). The ingredients in the PCF formulations were Cheddar cheese, butter, water, milk permeate powder, and LP (at a ratio of 2.0, 2.4, 2.8, 3.2, 4.0, 5.0, and 6.0%) were formulated to contain 17.0% protein, 25.0% fat, 44.0% moisture, and 2.0% salt. PCF made with 2.0% DSP was also produced as a control. The PCF was prepared by mixing all ingredients in a kitchen aid to make a homogeneous paste. A 25 g sample of the mixture was cooked in the rapid visco analyzer for 3 min at 95°C with 1000 rpm for the first 2 min and 160 rpm for the last min. The PCF was then transferred into molds and refrigerated till further analyses. The PCF was analyzed for moisture, pH, end apparent cooked viscosity, hardness, melted diameter, and melting temperature. The experiment was repeated 3 times using different batches of LP. The moisture of PCF ranged from 42.3 to 44.0% with a pH of 5.6 to 5.8. The end apparent cooked viscosity increased from 818.0 to 2060.0 cP as the level of LP raised from 0.63 to 1.90%, while it was 660.0 cP in control. The hardness of PCF made with LP elevated from 61.9 to 110.1 g as the level of LP increased; however, it was 85.6 g in control. The melted diameter decreased from 43 mm in control to 29 mm in 1.90% LP, while the melting temperature of PCF increased from 37.7°C in control to 59.0°C in 1.90% LP. We conclude that LP can be utilized as a substitute for DSP in PCF manufacture and has more capacity than DSP.

Keywords: Processed cheese food, Lactose-6-phosphate, Emulsifying salts, Functional properties, Protein
cessed cheese. In addition, PCF functionality may be classified into 3 broad categories: features linked to the rheology of raw cheese (fracture characteristics), cooking properties (flowability), and flavor/aroma properties (Guinee, 2011). The ingredients of PC include fat, moisture, intact casein, ES, and pH impact the functional properties of PCF.

ES, which consist of monovalent cation and polyvalent anions, are essential in making PCF. Different types of ES can be utilized in the manufacturing of PCF, namely, mono-, di-, trisodium phosphates, di-potassium phosphate, sodium hexametaphosphate, sodium acid pyrophosphate, sodium aluminum phosphate, sodium citrate, potassium citrate, sodium tartrate, and sodium-potassium tartrate. Trisodium citrate and sodium phosphate dibasic (DSP) are commonly used as ES in PCF manufacturing. ES play an important role in improving the emulsification characteristics of casein by sequestering calcium from the insoluble calcium-paracaseinate-phosphate network in natural cheese or the aggregated casein network in casein containing ingredients. As a result, the sequestered calcium partially disrupts the major molecular forces that cross-link the various casein monomers in the network. The protein is hydrated and dispersed because of this disruption. The partially dispersed monomers of casein have emulsification properties because they have hydrophilic and hydrophobic portions. The hydrophilic portion is linked to the aqueous phase, while the hydrophobic portion is linked to the fat phase, preventing oil separation in the presence of heating and mixing to produce a homogeneous product (Guinee, 2004; Hammam et al., 2022).

Lactose-6-phosphate (LP) is not certified to be utilized as an alternative to ES in making PC on a large scale. LP is naturally present in milk and milk products with low concentrations (Lifran et al., 2007; Thum et al., 2015). LP was initially identified in bovine milk (Cumar et al., 1965) and was recently identified in caprine milk (Albrecht et al., 2014). It presents in low concentrations, and due to the similar structure to lactose, it is hard to be fractionated and purify. LP is an organic molecule that replaces lactose’s hydrogen with monophosphate. A phosphate group is connected to the lactose galactose portion of most LP molecules. Approximately 90% of LP is coupled to galactose in pharmaceutical-grade lactose, while 10% is bound to glucose (Breg et al., 1988). LP is a chemical molecule generated from lactose that has the potential to behave as ES. It could reduce the amount of sodium and phosphate in PCF. High sodium intake in human nutrition is a risk factor for various disorders, notably causing high blood pressure and heart diseases (Wang and Labarthe, 2011).

Not enough literature on LP is available, and there is a lack of information to understand the composition and structure of this component. Additionally, there was no published data about using LP as ES in PCF. The limitations of studies on LP are as follow: i) Obtaining the right LP combination; ii) Finding pure lactose devoid of LP and other contaminants; iii) Directly analyzing LP and studying the emulsifying properties of LP. Thus, the study hypothesizes using LP as an alternative to ES in PCF manufacture. The main goals of this study were to prepare LP and produce PCF with different concentrations of LP instead of DSP and study the functional properties of the final product.

### MATERIALS AND METHODS

#### Lactose-6-phosphate preparation

LP was prepared using the methodology reported by Inoue et al. (2002) with some modifications. It was prepared by mixing 1 mol of α-D- (+) lactose monohydrate (C_{12}H_{22}O_{11}.H_{2}O; Fisher chemical; L5–500) with 0.5 mol of sodium cyclo-triphasosphate (Frontier Scientific; Cat # 343031). The pH of the recombined solution was adjusted using sodium hydroxide to get a pH of 12. The solution was stirred for 3 d at room temperature and then stored at 4°C for 24 h. The solution was separated into 2 layers. The top layer was removed using a syringe, and the bottom layer was processed further. The bottom layer was diluted with distilled water with a ratio of 1:2.2 into a 1000 mL glass Erlenmeyer flask. A 25 g

### Table 1. Mean (n = 3) relative abundance of lactose and lactose-6-phosphate (LP) for α-lactose monohydrate, and phosphorylation lactose (LP) using a mass spectrometer

<table>
<thead>
<tr>
<th>Treatments1</th>
<th>Lactose (Relative abundance % ± SD)</th>
<th>Lactose-6-phosphate (Relative abundance % ± SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>69.32 ± 16.87ª</td>
<td>0.89 ± 0.16ª</td>
</tr>
<tr>
<td>LP</td>
<td>15.58 ± 9.44ª</td>
<td>60.74 ± 38.89ª</td>
</tr>
</tbody>
</table>

Treatments1: Control = α-lactose monohydrate without any treatment; LP = The powder of phosphorylation lactose.

ªᵇ Means in the same row not sharing a common superscript are different at \( P < 0.05 \).
of activated carbon (Darco G60, 100 mesh, powder, Aldrich Chemical Company) was added to 250 mL of the lower portion of the solution, mixed well, and set for 5 min at room temperature. The mixture was filtered through filter paper (Cat No 1001 125, Whatman). The mix and filtration steps were repeated 7 times until having a transparent (colorless) solution. Then, the solution was evaporated using Hei-VAP Value laboratory vacuum evaporator (Heidolph Instruments GMbH & Co. KG) at 70°C, 350 mbar, and 100 rpm to obtain a recombined solution with 52% total solids (TS). Three different batches of recombined solutions were used in this study.

**Mass spectrometer**

The recombined solutions were dried before being analyzed with the mass spectrometer. The recombined solutions were freeze-dried using the methodology reported by Jouppila and Roos (1994) with some modifications. The recombined solutions were freeze-dried using a Labconco freeze-dryer (Lyph-Lock 6, Labconco, Kansas City, KS, USA) for 72h (temperature < −40°C; pressure < 0.1 mbar). For freeze drying, the solutions were poured into Petri dishes (15 mL in each to prepare samples) and frozen at −79°C using an ultralow freezer (NUAIRE −85 ± 1°C Ultralow Freezer) for 24h.

After drying the solutions, the mass spectrometer was performed using an Qtrap 5500 triple quadrupole mass spectrometer (AB Sciex, Foster City, CA, USA). Detection was achieved by electrospray ionization (ESI), operating in negative ion mode, with precursor ion scans performed with an m/z range of 300–450. Nitrogen (20 psi) was used as the curtain gas. The ion source was operated at −4500 V (V), a temperature of 300°C, and a pressure of 14 psi for nebulizer (GS1) gas. Default settings were used for the entrance potential of the collision cell (−10V), declustering potential (−90V), collision energy (−38V), and collision cell exit potential (−19V). Each sample was prepared in a concentration of 100 ng/mL (by weight) in water. Before analysis, the samples were filtered with a 4 mm, 0.2 um nylon syringe filter. Three different batches of recombined solutions were used in this study. A sample was applied as a control using α-lactose monohydrate.

**Processed cheese formulations**

The PCF formulations (44.0% water, 25.0% fat, 17.0% protein, and 2.0% salt) were prepared using TechWizard software (Metzger, 2010). The ingredients used in making PCF were Cheddar (Great Value, Mild Cheddar Cheese, Bentonville, AR), water, salt (Morton Salt, INC., Chicago, IL), salted butter (Land O Lakes Half Stick salted Butter, INC., Arden Hills, MN), deproteinized whey (Bondgrads’ Creameries, Perham, MN), and dibasic sodium phosphate (Fisher Scientific, Fair Lawn, New Jersey) as shown in Table 2.

Cheddar cheese, butter, water, and milk permeate powder were mixed using a KitchenAid 5KSM125BOB mixer was used (Artisan 4.8L Tilt-Head Stand Mixer). The mix time was for 30 min and speed at 6–7 to produce a homogenous paste. The mixture from Kitchen Aid was divided into 8 lots of 300 g. The first mix was used as a control (2% of DSP) (T1). The second (T2), third (T3), fourth (T4), fifth portion (T5), sixth portion (T6), seventh portion (T7), and eighth portion (T8) had 2.0, 2.4, 2.8, 3.2, 3.6, 4.0, 4.4, and 4.9% of recombined solutions. The level of LP was 0.63, 0.76, 0.88, 1.01, 1.26, 1.58, and 1.90% in T2, T3, T4, T5, T6, T7, and T8, respectively. For recombined solution, we prepared 200g in each trial that was 52% total solids.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Cheddar cheese</th>
<th>Water</th>
<th>Salted butter</th>
<th>Deproteinized whey</th>
<th>Salt</th>
<th>Sodium phosphate dibasic</th>
<th>Recombined solution</th>
<th>Amount of Lactose-6-phosphate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>66.95</td>
<td>18.10</td>
<td>3.45</td>
<td>7.45</td>
<td>2</td>
<td>2</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>T2</td>
<td>66.95</td>
<td>18.12</td>
<td>3.45</td>
<td>7.45</td>
<td>2</td>
<td>—</td>
<td>2</td>
<td>0.63</td>
</tr>
<tr>
<td>T3</td>
<td>67.01</td>
<td>18.05</td>
<td>3.43</td>
<td>7.10</td>
<td>2</td>
<td>2.4</td>
<td>—</td>
<td>0.76</td>
</tr>
<tr>
<td>T4</td>
<td>67.07</td>
<td>17.97</td>
<td>3.41</td>
<td>6.74</td>
<td>2</td>
<td>2.8</td>
<td>—</td>
<td>0.88</td>
</tr>
<tr>
<td>T5</td>
<td>67.13</td>
<td>17.89</td>
<td>3.38</td>
<td>6.38</td>
<td>2</td>
<td>3.2</td>
<td>4</td>
<td>1.01</td>
</tr>
<tr>
<td>T6</td>
<td>67.24</td>
<td>17.74</td>
<td>3.33</td>
<td>5.67</td>
<td>2</td>
<td>3.8</td>
<td>4</td>
<td>1.26</td>
</tr>
<tr>
<td>T7</td>
<td>67.39</td>
<td>17.57</td>
<td>3.28</td>
<td>4.77</td>
<td>2</td>
<td>5</td>
<td>4</td>
<td>1.58</td>
</tr>
<tr>
<td>T8</td>
<td>67.46</td>
<td>16.93</td>
<td>3.25</td>
<td>4.34</td>
<td>2</td>
<td>6</td>
<td>4</td>
<td>1.90</td>
</tr>
</tbody>
</table>

1Treatments: Control = Processed cheese made with sodium phosphate dibasic; T2 = Processed cheese made with 0.63% lactose-6-phosphate; T3 = Processed cheese made with 0.76% lactose-6-phosphate; T4 = Processed cheese made with 0.88% lactose-6-phosphate; T5 = Processed cheese made with 1.01% lactose-6-phosphate; T6 = Processed cheese made with 1.26% lactose-6-phosphate; T7 = Processed cheese made with 1.58% lactose-6-phosphate; and T8 = Processed cheese made with 1.90% lactose-6-phosphate.
Cooking the processed cheese

The PCF formulations were cooked in the rapid visco analyzer (RVA) as described in other studies (Metzger et al., 2002; Kapoor et al., 2004; Kapoor and Metzger, 2005; Hammam et al., 2022). A total of 25 g of the mix with either DSP or recombined solution were transferred to a canister and then tempered at 40°C/15 min in a water bath (SWB-20L-3; Major Science, USA). The pH was adjusted between 5.6 to 5.7 using sodium hydroxide 40% wt/wt (Fisher Scientific, S318–1). The canister was cooked in the RVA (Perten RVA 4500, Macquarie Park NSW 2113, Australia). The canister was cooked at 95°C for 3 min at a speed of 1000 rpm for the first 2 min and 160 rpm during the last min. The end apparent cooked viscosity of PCF was measured at 95°C at the end of the cooking time by calculating the mean of the last 5 viscosity values, as shown in Figure 1. PCF was then poured into copper cylinders (20 mm diameter × 30 mm height) and plastic molds (28.3 mm diameter × 25 mm height) sealed with aluminum foil. PCF was refrigerated till further analyses. This experiment was repeated 3 times.

Compositional analyses

The final PCF was analyzed for moisture using a forced draft oven (AOAC, 2000, method 990.20; 33.2.44) and pH (Hanna Instruments, model HI2202).

Functional analyses

Dynamic Stress Rheometer (DSR). The dynamic rheological analysis (DSR) determined the initial melt characteristics of PCF and indicated molecular interactions. The melting point is the lowest temperature, where a material shifts from primarily elastic to primarily viscous (Prow and Metzger, 2005). DSR was done as described in a previous study (Hammam et al., 2022) using a rheometer (MSR 92, Anton Paar, Graz, Austria) equipped with a 25-mm parallel plate geometry. The cheese was cut into 2 mm thick slices at room temperature using a wire cutter. A stress sweep test for PCF was performed at a frequency of 1.5 Hz and a range of 1 to 1000 Pa stress at 20°C using the rheometer with parallel plate geometry. The stress sweep test found that the maximum stress limit for the linear viscoelastic region was 500 Pa.
The melting temperature of PCF was determined using the heat rate of 1°C/min. The temperature ranged from 20 to 90°C using a frequency of 1.5 Hz and stress of 500 Pa (linear viscoelastic region). Elastic modulus ($G'$), viscous modulus ($G''$), and tangent angle ($\tan \delta$). The temperature at which $\tan \delta = 1$ ($G''/ G'$) was known as the cheese melt temperature. DSR was done in triplicate.

**Schreiber melt test.** The melt of PCF was done using the Schreiber melt test. PCF with 28.5 mm diameter was removed from a mold and cut using a wire cutter to obtain 7 mm high. PCF was transferred to Petri dishes and left at 90°C for 7 min. The diameter of the melted cheese was measured in four different spots using a ruler after cooling and reported in mm.

**Texture profile analysis (TPA).** The hardness of the PCF was measured using the texture profile analyzer (TPA) as described by (Hammam et al., 2022). A 20 mm high PCF with a 20 mm diameter was cut using a wire cutter and transferred to the texture analyzer (TA.XT-Plus, 6 Patton Drive, South Hamilton, MA). TPA was performed using uniaxial double bite compression (50-mm diameter cylindrical flat probe, 10% compression, and one mm/s crosshead speed). A 20 mm high PCF with a 20 mm diameter was transferred to the TPA. The hardness of PCF was referred to as the top of the first force that results from the first compression. TPA was performed on 2 samples of each replicate.

**Statistical Analysis**

Statistical analysis was performed to study the concentration of LP on PCF’s functional properties. The ANOVA test was done using R software (R × 64–3.3.3, R Foundation for Statistical Computing). Mean separation was done using the least significant difference (LSD) test at $P < 0.05$.

**RESULTS AND DISCUSSIONS**

**LP determination**

Table 1 presents the relative abundance of lactose for control and the recombined solutions, which were 69.32, and 15.58%, respectively. The amount of lactose significantly dropped ($P < 0.05$) for control and the recombined solutions, which is expected due to the chemical reaction between the monosaccharides and hydrolyzed disaccharides with sodium hydroxide (Zheng et al., 2015). In addition, the degradation of lactose under alkaline conditions could be another reason for the reduction of lactose (Lewkowski, 2001). A study was done on hemicellulose sugar shows that the alkaline conditions release acetyl and acidic groups and organic acids that lead to reducing of the pH (Nikzad et al., 2014).

LP was detected in all treatments with various levels. Previous studies have found that the LP was presented at lower concentrations in lactose and whey, 368.0 and 0.70 ppm, respectively (Lifran, 2007; Thum et al., 2015). The relative abundance of LP was 0.89, and 60.74% for control, and the recombined solutions, respectively. The recombined solutions had a higher ($P < 0.05$) LP level than the control. Similar results were found in Inoue et al. (2002), who phosphorylated lactose to obtain 33.0% LP.

**Composition of processed cheese**

The mean composition of moisture of PCF is illustrated in Table 3. The ANOVA with MS and $P$-values for moisture of the PCF is exhibited in Table 4. The moisture of PCF in control, T2, T3, T4, T5, T6, T7, and T8 were 44.0, 42.8, 43.7, 43.6, 43.1, 42.3, and 43.1%, respectively. The low moisture content in the final PCF of the LP compared with the targeted moisture content (44.0%) could be related to differences in TS of the recombined solutions that were used in this study, which resulted in decreasing the moisture content in the final PCF. No significant difference ($P > 0.05$) was detected in PCF moisture between control and PCF treated with different concentrations of LP. The moisture of treatments ranged from 42.3 to 44.0%, which is within the expected level for PCF.

The mean composition of PCF’s pH is shown in Table 3. The ANOVA with MS and $P$-values for pH of the PCF is presented in Table 4. The pH of PCF in control, T2, T3, T4, T5, T6, T7, and T8 were 5.6, 5.6, 5.7, 5.7, 5.7, 5.7, 5.7, and 5.8, respectively. The addition of 1.90% LP resulted in the highest pH ($P < 0.05$) in the final PCF compared with other treatments. This could be related to the high sodium level in the recombined solutions that was noticeable at 1.90% LP. However, no significant difference ($P > 0.05$) was detected in the pH between control and PCF treated with up to 1.58% of LP. In this study, PCF’s pH ranged from 5.6 to 5.8, which was in the typical range (5.4–5.8) of pH of PCF (Palmer and Sly, 1943; Caric et al., 1985; Marchesseau et al., 1997; Kapoor and Metzger, 2008). When the pH of the PCF is less than 5.4 or more than 5.8, the stability of PCF emulsion is reduced (Palmer and Sly, 1943). Similar results have been found for some PCF with different concentrations of ES (Gupta et al., 1984). Since the main role of ES is calcium sequestration and pH adjustment, which increase with increase the amount of ES. These interactions improve the final product’s homogenization by hydrating caseins and increasing
the interaction between water and fat phases (Kapoor and Metzger, 2008). The slight differences in pH could affect the viscosity of PCF (Caric et al., 1985).

**Functional Properties**

*End apparent cooked viscosity.* The mean values of cooked viscosity (cP) of PCF measured using the RVA are exemplified in Table 5. The ANOVA with MS and P-values for cooked viscosity of the PCF is illustrated in Table 6. The viscosity of PCF in control, T2, T3, T4, T5, T6, T7, and T8 were 660.5, 818.9, 1065.3, 1205.3, 1175.2, 1691.9, and 2060.5 cP, respectively. There was a significant difference ($P < 0.05$) in the end apparent cooked viscosity of PCF made with different levels of the LP. This could be due to the differences in the amount of sodium coming from the recombined solutions that affect the pH. The variations in the pH of the final PCF might impact the end apparent cooked viscosity of PCF. When the pH of PCF elevated, the net negative charges of casein increased, and this, in turn, led to increases in the calcium-mediated cross-linking of casein molecules in the PCF gel network. When PCF is heated, the calcium-induced cross-linking of casein molecules restricts the movement of the casein chains, reducing flowability and increasing viscosity (Kapoor and Metzger, 2008; Marchesseau et al., 1997).

Similar results were reported by Kapoor (2007), who noticed increasing the viscosity of PCF with increasing pH to 5.8.

**Hardness.** The mean values of hardness (g) of PCF determined using the TPA are revealed in Table 5. The ANOVA with MS and P-values for the hardness of the PCF is displayed in Table 6. The hardness of PCF in control, T2, T3, T4, T5, T6, T7, and T8 was 85.6, 64.1, 66.3, 61.9, 72.4, 67.4, 77.8, and 110.1 g, respectively. Significant differences ($P < 0.05$) were detected in the hardness of PCF made with different levels of the LP. No significant difference ($P > 0.05$) was detected between PCF made with 2% DSP (control) and PCF treated with up to 1.58% of LP. The highest hardness was found in PCF made with 1.90% LP, which could relate to the pH of the final PCF. PCF’s pH significantly impacts its rheology, texture, and microstructure characteristics (Mulsow et al., 2007). As the pH of PCF increases, the net negative charges on caseins and the electrostatic repulsion in the casein matrix elevate. Higher repulsion at high pH should result in a more open and loose PCF network with improved water binding capacity and emulsifying ability during PCF manufacturing (Guinee et al., 2004). When increasing the pH of PCF, the hydrophobic interactions between individual casein molecules should decrease due to the increase in electrostatic repulsion (Horne, 1998; Lucey et al., 2003). This results in stronger hydrogen bonds and more calcium-mediated cross-links between casein molecules in PCF, which improves the strength of the PCF gel network and hence the hardness of the final PCF (Bulut-Solak and Akin, 2019). Another study found that the final pH of PCF affected its firmness (Templeton and Sommer, 1932; Marchesseau et al., 1997). They discovered that as the PCF’s pH increased from 5.0 to 6.2, its firmness improved up to around pH 5.8 (where it had the most firmness), but the firmness started to drop as the pH increased further from 5.8 to 6.2.

**Schreiber melt test.** The mean values of the Schreiber melt test of PCF made with the LP are presented in Table 7. The ANOVA with MS and P-values for the Schreiber melt test (mm) of the PCF is
Table 5. Mean (n = 3) viscosity and hardness (g) of processed cheese (PCF) with either lactose-6-phosphate or sodium phosphate dibasic (DSP) and 2% DSP

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Control</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>T5</th>
<th>T6</th>
<th>T7</th>
<th>T8</th>
</tr>
</thead>
<tbody>
<tr>
<td>Viscosity (cP)</td>
<td></td>
<td>660.5c</td>
<td>818.9de</td>
<td>1065.3cd</td>
<td>1205.3c</td>
<td>1152.9c</td>
<td>1175.2c</td>
<td>1691.9b</td>
</tr>
<tr>
<td>Hardness (g)</td>
<td></td>
<td>85.6ab</td>
<td>64.1b</td>
<td>66.3b</td>
<td>61.9b</td>
<td>72.4ab</td>
<td>67.4b</td>
<td>77.8ab</td>
</tr>
</tbody>
</table>

1Treatments: Control = Processed cheese made with sodium phosphate dibasic; T2 = Processed cheese made with 0.63% lactose-6-phosphate; T3 = Processed cheese made with 0.76% lactose-6-phosphate; T4 = Processed cheese made with 0.88% lactose-6-phosphate; T5 = Processed cheese made with 1.01% lactose-6-phosphate; T6 = Processed cheese made with 1.26% lactose-6-phosphate; T7 = Processed cheese made with 1.58% lactose-6-phosphate; and T8 = Processed cheese made with 1.90% lactose-6-phosphate.

Means in the same row not sharing a common superscript are different at P < 0.05.

Table 6. Mean squares and P-values for end apparent cooked viscosity and hardness of process cheese food (PCF) with either lactose-6-phosphate or disodium phosphate (DSP)

<table>
<thead>
<tr>
<th>Factor</th>
<th>df</th>
<th>End apparent cooked viscosity</th>
<th>Hardness</th>
</tr>
</thead>
<tbody>
<tr>
<td>Replication</td>
<td>2</td>
<td>230326 (&lt;0.05)</td>
<td>578.14 (0.32)</td>
</tr>
<tr>
<td>Treatment</td>
<td>7</td>
<td>61481 (&lt;0.05)</td>
<td>761.27 (0.21)</td>
</tr>
<tr>
<td>Error</td>
<td>14</td>
<td>27724</td>
<td>467.38</td>
</tr>
</tbody>
</table>

Lactose-6-phosphate (LP) was prepared and used successfully as an alternative to DSP in making PCF. When comparing PCF at the same pH and TS, we assume that the higher the degree of casein dispersion caused by using a certain type of ES during cooking, the firmer the product after cooling. This study concluded that as LP concentration elevated, the pH increased, which had a significant impact on PCF properties. This study determined that LP could replace DSP to produce PCF with less melt than DSP. The capacity of LP was demonstrated to be greater than that of DSP as ES in PCF. The concentration of LP can be adjusted based on the required functional characteristics of PCF. The level of LP has a significant impact on the physical and chemical properties of PCF. With the lowest level of LP in T2, we did not find any oiling off. The study found that the higher the LP, the more significant viscosity, hardness, melt temperature, and lower melting area. The next step of future study could be making LP with milk permeate calcium (Shirashoji et al., 2016) and low pH due to the poor emulsification of fat and water at low pH. Also, this may be related to the level and buffering capacity of ES used on PCF, which plays a critical role in the melting points of PCF (Marchesseau et al., 1997).

**Effect of LP in the functionalities of PCF.** Table 2 shows the amount of LP in each treatment. LP was 0.63, 0.76, 0.88, 1.01, 1.26, 1.58, and 1.90% in T2, T3, T4, T5, T6, T7, and T8. The amount of phosphate was 1.32% in control which was between T6 and T7. Comparing the properties of PCF between control and the lowest level of phosphate that was in T2 shown in Table 2. There was no significant difference in terms of viscosity, hardness, and melting temperature of PCF. There was a significant difference in the Scheiber melt test. T2 had less melting area compared with the control. The capacity of LP was shown to be more compared with DSP. LP has improved protein heat stability, better emulsifying properties and solubility (Julmohammad, 2017).

**CONCLUSIONS**

LP was prepared and used successfully as an alternative to DSP in making PCF. When comparing PCF at the same pH and TS, we assume that the higher the degree of casein dispersion caused by using a certain type of ES during cooking, the firmer the product after cooling. This study concluded that as LP concentration elevated, the pH increased, which had a significant impact on PCF properties. This study determined that LP could replace DSP to produce PCF with less melt than DSP. The capacity of LP was demonstrated to be greater than that of DSP as ES in PCF. The concentration of LP can be adjusted based on the required functional characteristics of PCF. The level of LP has a significant impact on the physical and chemical properties of PCF. With the lowest level of LP in T2, we did not find any oiling off. The study found that the higher the LP, the more significant viscosity, hardness, melt temperature, and lower melting area. The next step of future study could be making LP with milk permeate...
Treatments: Control = Processed cheese made with sodium phosphate dibasic; T2 = Processed cheese made with 0.63% lactose-6-phosphate; 1.58% lactose-6-phosphate; and T8 = Processed cheese made with 1.90% lactose-6-phosphate.

Table 7. Mean (n = 3) Schreiber melt test (mm) and melting temperature (°C) of processed cheese (PCF) with different levels of lactose-6-phosphate

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Control</th>
<th>T2</th>
<th>T3</th>
<th>T4</th>
<th>T5</th>
<th>T6</th>
<th>T7</th>
<th>T8</th>
</tr>
</thead>
<tbody>
<tr>
<td>Schreiber melt test (mm)</td>
<td>43.2a</td>
<td>34.2b</td>
<td>30.5c</td>
<td>32.5b</td>
<td>31.1b</td>
<td>30.4bc</td>
<td>32.6bcd</td>
<td>29.3d</td>
</tr>
<tr>
<td>Melting temperature (°C)</td>
<td>37.7</td>
<td>43.0a</td>
<td>51.1b</td>
<td>53.9a</td>
<td>49.1cd</td>
<td>55.4bc</td>
<td>57.9b</td>
<td>58.9a</td>
</tr>
</tbody>
</table>

a–d Means in the same row not sharing a common superscript are different at P < 0.05.

ACKNOWLEDGMENTS

This work has been made possible through financial support from Midwest Dairy Foods Research Center (St. Paul, MN) and Qassim University.

REFERENCES


Table 8. Mean squares and P-values of Schreiber melt test (mm) and melting temperature (°C) of process cheese food (PCF) with either lactose-6-phosphate or disodium phosphate (DSP)

<table>
<thead>
<tr>
<th>Factor</th>
<th>df</th>
<th>Diameter</th>
<th>DSR</th>
</tr>
</thead>
<tbody>
<tr>
<td>Replication</td>
<td>2</td>
<td>0.849 (0.76)</td>
<td>6.56 (0.67)</td>
</tr>
<tr>
<td>Treatment</td>
<td>7</td>
<td>59.19 (&lt;0.05)</td>
<td>161.81 (&lt;0.05)</td>
</tr>
<tr>
<td>Error</td>
<td>14</td>
<td>3.001</td>
<td>16.43</td>
</tr>
</tbody>
</table>

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